Cleaning, Milling, and Baking Tests with Hard Red Winter Wheat Containing Deoxynivalenol[1]

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ABSTRACT

Seven commercial lots of 1982 hard red winter wheat were selected for cleaning, milling, and baking tests. Five scab-infected lots had deoxynivalenol (DON) concentrations ranging from 0.64 to 5.10 ppm, and two lots were scabfree controls. Each lot was cleaned by four methods: 1) a normal, single cleaning to obtain maximum screenings with a minimum of wheat lost; 2) double cleaning using the normal flow; 3) single cleaning with suction increased on the millerator and the entoleter aspirator; and 4) single cleaning followed by washing the wheat with water. None of the cleaning methods completely removed DON, and the special methods showed little or no advantage over the normal, single cleaning. Cleaning efficiency varied with DON concentration among lots of cleaned wheats (means of four cleaning methods) ranging from 48 to 86% of that in uncleaned wheat. The cleaned wheats were milled with a Miag Multomat mill. All mill fractions from scab-infected wheat contained DON. Concentrations of DON were generally lower in flours, and higher in offals, than in the cleaned wheat. Average DON concentration of straight-grade flour was 44 and 75% of that in uncleaned and cleaned wheat, respectively. DON was not destroyed by baking bread. Only bread from flours with highest levels of DON had slightly reduced loaf volumes and off-colors in bread crumb compared to controls.

During maturation of the 1982 hard red winter (HRW) wheat crop, wet weather in parts of the midwestern United States caused an unusually high occurrence of scab, a disease caused by the fungus *Fusarium graminearum* (Weise 1977). Occurrence of 4-deoxynivalenol (DON), a metabolite of *F. graminearum*, was also common in the scabby wheat (Bertelsen 1982, Kansas Agricultural Experiment Station 1983). Possible DON contamination of wheat-based foods was a matter of concern (Anonymous 1982), because DON had been reported to exhibit various toxicological effects in animals (Ueno 1983). Therefore, this study was initiated in regard to HRW wheat from the midwestern United States to determine: 1) whether seed cleaners could effectively remove or significantly reduce scab-infected wheat, 2) how DON would be distributed among various mill products, and 3) how various degrees of scab infection might affect the baking properties of flour. Results have been reported from milling and baking tests with various wheats from eastern Canada (Scott et al 1983, 1984; Young et al 1984) and from milling tests with soft wheats from the United States (Hart and Braselton 1983, Seitz et al 1985). Previously, Finney (1954) reported on the physical, chemical, and baking properties of flour milled from scabby HRW wheat.

MATERIALS AND METHODS

Wheat Lots

All wheat used for this study was from the 1982 crop grown commercially in northeastern and north central Kansas. Five scab-infected and two control lots were cleaned and milled by the Department of Grain Science and Industry, Kansas State University (Table I). Lot 2 was formed by blending equal amounts of lots 3 and 6. Grades and percentages of scab (assessed visually by special request) were determined by the Federal Grain Inspection Service, USDA. Each lot was thoroughly blended before sublots were removed for cleaning tests. A representative sample (about 5 kg) for chemical analyses was obtained by blending several subsamples removed from different locations within the blended lot.

Cleaning Tests

Wheat from each lot was cleaned by four methods, with 20 bu (540 kg) used for each method. Samples (about 5 kg) of cleaned wheat for chemical analyses were formed by blending several portions taken from the wheat stream exiting the cleaner. After screenings were weighed and blended, about 500 g was removed for chemical analyses. The cleaning methods were as follows:

Normal. The wheat was cleaned at the rate of 60 bu (1,620 kg)/hr using the cleaning house shown in Figure 1. Equipment settings were such that maximum screenings were removed with a minimum of wheat lost. The Superior cylinder separators were used instead of the Simon Carter disk separators and Simon Carter precision width grader.

Double. The wheat was cleaned twice in the cleaning house using the normal flow.

High aspiration. Using the normal cleaning flow, the suction was increased on the Simon Carter millerator and entoleter scourer aspirator. All high-aspiration runs used the same air settings.

Washer. After a single pass through the normal cleaning flow, the wheat was washed in a Smico wheat washer. Wheat flow rate was 60 bu/hr, and the water rate was adjusted so that the wheat was thoroughly wetted. The wheat moisture after washing was 14–15%.

Milling Tests

Cleaned wheat (81 kg) was tempered to 16% moisture, then milled on a Miag Multomat mill having the configuration shown in Figure 2. After each mill fraction was weighed and blended, about 300 g was taken for chemical analyses.

Additional flours from lots 4 and 5 were obtained by using an Allis-Chalmers experimental mill (straight grade) and a Weber hammer mill (whole wheat) at the U.S. Grain Marketing Research Laboratory.

Baking Tests

Baking qualities of flours were evaluated by using an optimized breadmaking method of Finney (1984). One 100-g loaf was baked from each of the 28 straight-grade flours (7 lots, four cleaning methods) from the Miag Multomat mill. After being graded, each loaf was sliced, air-dried, and ground in preparation for DON analyses.

To determine DON contents of bread crumb versus crust, straight-grade and whole wheat flours from lots 4 and 5 were used. One 100-g loaf was baked from each flour. Each loaf was sliced, crumb immediately separated from crust by hand, and the portions air-dried and ground.

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Analytical

About 300 g of each whole wheat sample was ground to about 20 mesh. Wheat was fed through the grinder (Falling Number, KT-30, Kansas City, MO) in small portions (30–50 g) with each portion taken after remixing the unground sample to maintain homogeneity. Similarly, samples used for extraction were always prepared by combining several portions from a reblended ground sample.

For determining DON in ground whole wheats, mill fractions, and bread, the method of Scott et al (1981) with minor modifications was used, except that high-pressure liquid chromatography (HPLC) replaced gas chromatography for detecting DON in final extracts. The methodology has been described previously (Seitz and Bechtel 1985). Limit of detection was about 0.01 ppm. Because the number of samples to be analyzed was large (476), a representative sample (50 g) of each grain or milled product was extracted only once, but each final cleaned extract was analyzed twice by HPLC. A sample was reanalyzed (extraction, cleanup, and HPLC) if there was good reason to question the accuracy of the result from the first analysis.

Protein, ash, and moisture were determined by AACC methods 46-10, 08-01, and 44-15A, respectively (AACC 1976). The ash method was modified such that samples were placed in the muffle furnace at 427° C (800° F), ignited, then heated at 496° C (925° F) for 16 hr and finally at 538° C (1,000° F) for 4 hr.

Data Processing

To obtain an overall picture of the distribution of DON among mill fractions the data was processed as follows: For each wheat milled, DON concentrations in mill stream products were expressed as percent DON concentration in the cleaned wheat. Then for each fraction, mean DON concentrations were calculated and plotted (Figs. 3–5). Protein and ash data were processed in the same way.

RESULTS AND DISCUSSION

Cleaning Tests

None of the four cleaning methods was particularly effective for DON removal (Table II). From method averages (Table II), and from amounts of screenings and their DON concentrations (Table

TABLE I
Deoxynivalenol (DON) and Scab in 1982 Hard Red Winter Wheats Used for Cleaning and Milling Tests

Lot	DON (ppm)	Scab (%)	Test Wt (lb/bu)	Grade	Protein (%)
1	0.64	1.9	58.4	2	12.2
2	0.79	1.2	59.0	2	11.9
3	1.25	2.4	56.0	2	12.0
4	1.49	2.9	56.7	2	12.6
5	5.10	12.8	53.2	5	13.2
6	0	0	61.9	1	11.6
7	0	0	59.6	1	12.9

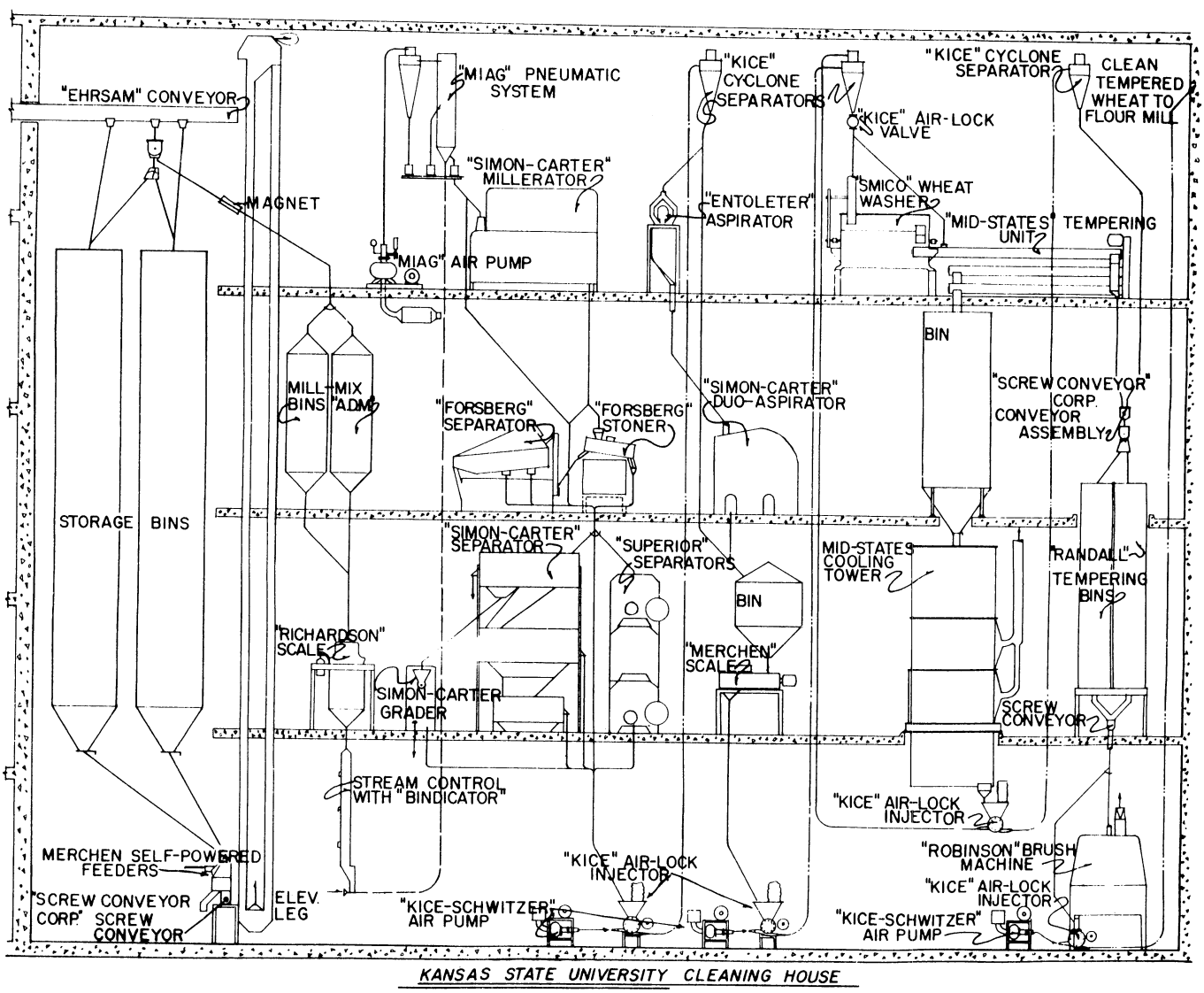

Fig. 1. Flow diagram for Kansas State University cleaning house.

III), it is evident that the double cleaning method generally gave the cleanest wheat. However, differences among results of the methods were small, so there appeared to be little or no advantage to using any method other than normal cleaning.

The presence of light-weight, severely infected kernels or pieces of kernels caused screenings to have high DON concentrations (Table III). Even the two control lots (6 and 7), which appeared scabfree and had no detectable DON in bulk samples, had measurable amounts of DON in screenings (Table III).

Degree of DON removal varied considerably among lots as indicated by lot means shown in Table II. The DON in lot 1 wheat was apparently distributed fairly evenly among lightly infected kernels of near-normal size and weight that could not be removed by the cleaner. In lot 3 wheat, however, a larger proportion of the DON was apparently contained in light-weight, severely infected kernels that could be removed by the cleaner. Because cleaning efficiencies for lots 2 and 3 were similar, blending of scabby and scabfree lots had no effect on efficiency for removal of DON. Chemical, physical, and microscopic studies of scab-infected HRW wheat have shown that degree of *Fusarium* infection varied substantially among kernels, and that kernels of nearly normal size and weight had significant fungal invasion and DON contents (Seitz and Bechtel 1985). Tests with eastern Canadian wheats (Scott et al 1983, 1984; Young et al 1984) and with soft wheats from Missouri and Ohio (Seitz et al 1985) also showed that cleaners did not efficiently remove DON.

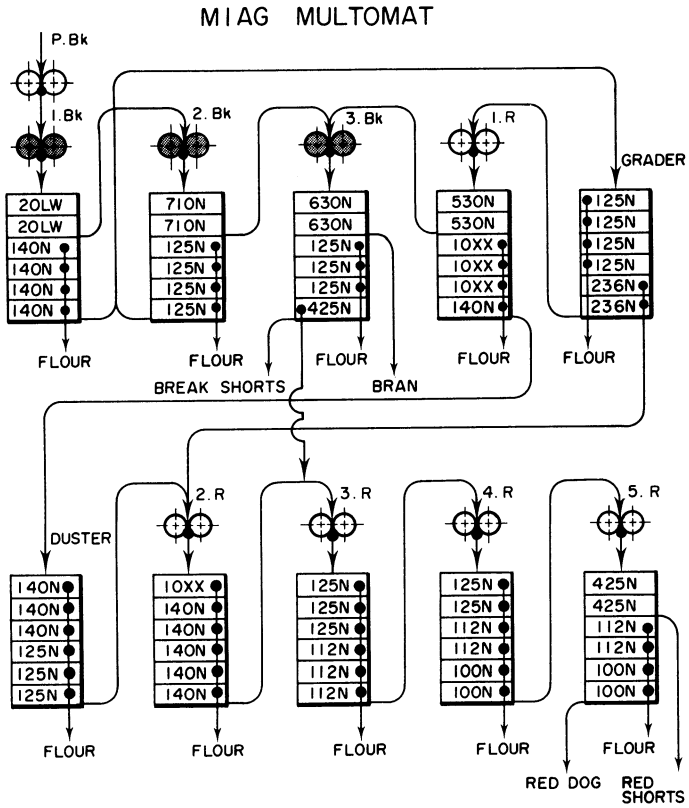

Fig. 2. Flow diagram for Miag Multomat mill.

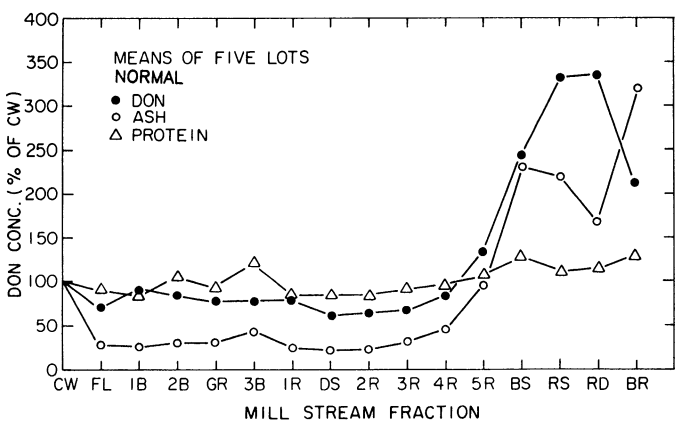

Fig. 3. Deoxynivalenol (DON) (•), ash (o), and protein (Δ) contents of hard red winter wheat mill fractions expressed as percent of the respective component in wheat cleaned by the normal method (Materials and Methods). Fractions include cleaned wheat (**CW**), straight-grade flour (**FL**), break flours (**1B, 2B, 3B**), grader (**GR**), reduction flours (**1R, 2R, 3R, 4R, 5R**), duster (**DS**), break shorts (**BS**), reduction shorts (**RS**), red dog (**RD**), and bran (**BR**). All values are means of five lots. Means and ranges of coefficients of variation (%) for DON, ash, and protein were: 34.2, 22–60; 18.1, 4.0–30.7; and 2.9, 1.0–3.7, respectively.

Milling Tests

Yields of fractions from the Miag Multomat mill were within normal ranges for all lots, and no consistent trends or unusual changes in yields were caused by differences in degree of scab infection or method of cleaning. Straight-grade flour yields (means of four cleaning methods) for lots 1–7, in order, were: 69.7, 73.4, 69.5, 68.6, 68.1, 71.0, and 72.1%; overall mean yield was 70.3% (SD 2.2). Also, plots of ash and protein contents versus mill stream

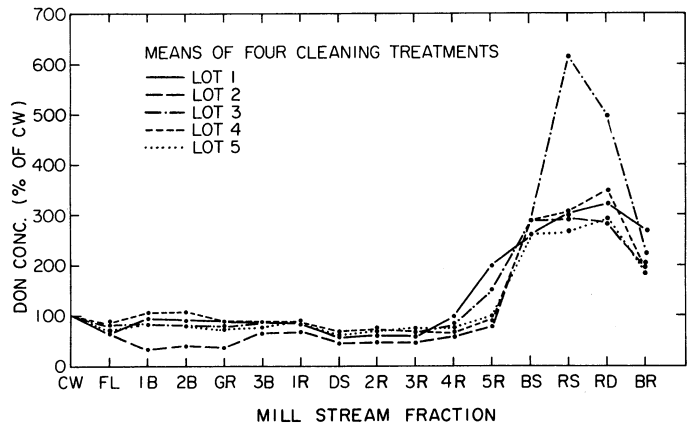

Fig. 4. Lot-to-lot comparison of deoxynivalenol (DON) contents of hard red winter wheat mill fractions. Values are expressed as percent of the DON concentration in cleaned wheat and represent means of four cleaning methods. Fractions are identified in Fig. 2. Means and ranges of coefficients of variation for lots 1–5 were, in order: 17.2, 4–29; 33.6, 18–48; 21.4, 15–35; 14.0, 2–38; and 30.5, 10–48.

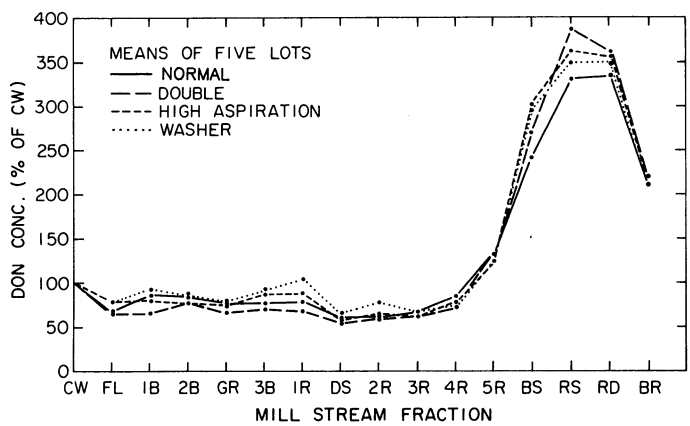

Fig. 5. Deoxynivalenol (DON) contents of fractions from the milling of hard red winter wheats cleaned by four methods. Values are expressed as percent of the DON concentration in cleaned wheat and represent means of five lots. Fractions are identified in Fig. 2. Means and ranges of coefficients of variation for normal, double, high aspiration, and washer methods were: 34.2, 22–60; 36.9, 15–54; 24.9, 11–44; and 29.3, 11–60, respectively.

fraction (typical curves shown in Fig. 3) did not indicate that gross mill stream yields were affected by scab infection or cleaning method.

DON was found in all mill fractions of DON-contaminated lots (Figs. 3–5), but not in any from control lots. Mean DON concentrations in straight-grade flours from wheats cleaned by the four methods could not be considered different because of the relatively large standard deviations (Table IV). However, certain trends in the distribution of DON among mill stream fractions suggest that the concentration of DON in flours was affected slightly by cleaning method. Overall means for DON concentrations in straight-grade flours were 44 and 72% of that in uncleaned and cleaned wheats, respectively. Concentrations of DON in straight-grade flours and cleaned wheats were highly correlated ($r = 0.96$, $n = 20$, $P < 0.001$).

Generally, flour fractions had lower, and offal fractions higher, DON concentrations than cleaned wheat (Figs. 3–5). This distribution pattern was similar among lots, regardless of the DON content of the whole wheat or type of cleaning method, as indicated by generally high correlation coefficients (Mostly 0.82–0.98, $P < 0.001$). However, some differences among lots were apparent in plots of DON concentrations versus mill stream fraction (Fig. 4). The difference between lots 2 and 3 is an enigma, because lot 2 is a blend of lots 3 and 6. Whether or not the consistently low DON concentrations in lot 2 flours were caused by the blending is not known. It is also not clear why DON concentrations were unusually high in reduction shorts and red dog fractions from lot 3 wheat. Ash and protein data did not show differences among lots that would explain the unusual DON distribution for lot 3.

Effect of cleaning method on the distribution of DON among mill stream fractions is shown in Figure 5. DON concentrations were, in general, lowest in flours from double-cleaned and highest in washer-cleaned wheat. The difference was significant at $P \sim 0.014$ indicated by a two-tailed, paired-sample t test analysis of data pairs (double versus washer) from all flour fractions except the low-yield fourth and fifth reduction flours. Relatively high DON concentrations of first and second reduction flours from washer-cleaned wheat were noteworthy, because a similar pattern was prominent in flours from the milling of soft wheats (Seitz et al 1985). Method of cleaning had no apparent effect on distribution of ash and protein among mill stream fractions; i.e., plots for double-, high-aspiration, and washer-cleaned wheats were essentially identical to those shown in Figure 3 for normal cleaned wheat. Offal fractions, and fourth and fifth reduction fractions from washer-cleaned wheat had slightly reduced ash contents, apparently caused by minerals leaching into the wash water.

Our results, as well as results from other tests (Scott et al 1983, 1984; Seitz et al 1985; Young et al 1984) indicating a fractionation of DON among all mill fractions, are consistent with recent findings concerning the distribution of the fungus in infected kernels. Microscopical results demonstrated that the fungus showed a preference to aleurone and pericarp tissues, but hyphae were found throughout the entire endosperm tissue (Bechtel et al 1985). Offal fractions are composed mostly of aleurone and pericarp tissues (high ash and protein) whereas flour fractions are composed of mostly endosperm tissue (low ash and protein). By using the data in Figure 3 for normal cleaned wheat, we found that DON correlated slightly less with protein ($r = 0.69$, $P < 0.005$), and about the same with ash ($r = 0.84$, $P < 0.001$), as protein and ash did with each other ($r = 0.82$, $P < 0.001$).

Mill fractions from wheats cleaned by the other three methods gave essentially the same correlations. Also, these correlations were similar to those found for mill fractions from a group of soft wheats (Seitz et al 1985). Young et al (1984) reported a positive correlation ($r = 0.80$) between ergosterol and DON in flour and offal fractions, which indicated that the DON was associated with fungal growth rather than transported from the outer to the inner part of the kernel.

The DON contents of some of the flour fractions were unexpected. High-ash flours, which contain more aleurone and subaleurone tissues than low-ash flours, could reasonably be expected to have higher DON concentrations than low-ash flours. This expectation was generally met by flours from the third, fourth, and fifth reduction rolls (Figs. 2–4). However, with most low-ash flours, particularly those from the first and second break rolls and the first reduction roll, higher than expected DON concentrations were usually observed. These findings may have resulted from changes in kernel endosperm fracturing characteristics caused by the fungus. Microscopical studies have shown that the fungus

TABLE II
Deoxynivalenol Concentrations of Cleaned Wheats Expressed as Percent of Concentration in Uncleaned Wheats

Lot	Normal	Double	High Aspiration	Washer	Lot Mean	SD
1	78	88	88	89	86	5
2	62	43	34	52	48	12
3	46	41	61	45	48	9
4	66	69	72	74	70	4
5	53	36	56	72	54	15
Method mean	61	55	62	66		
SD	12	22	20	18		

TABLE III
Amounts and Deoxynivalenol (DON) Concentrations of Screenings Removed from 1982 Hard Red Winter Wheats Cleaned by Three Methods

Lot	Cleaning Method	Amount (% of Uncleaned Wheat)	DON Concentration (ppm)	DON Concentration (% of Uncleaned Wheat)
1	Normal	1.0	6.8	1,100
	Double	2.0	11.8	1,840
	High Asp.[a]	1.4	7.2	1,100
2	Normal	1.6	7.2	910
	Double	3.5	6.1	770
	High Asp.	3.3	8.1	1,000
3	Normal	2.5	7.8	620
	Double	4.5	10.8	860
	High Asp.	3.4	7.5	600
4	Normal	1.7	11.2	752
	Double	3.0	22.5	1,510
	High Asp.	2.5	14.2	953
5	Normal	5.7	18.0	350
	Double	8.0	15.3	300
	High Asp.	8.7	15.0	294
6	Normal	1.0	0.9	...
	Double	1.6	0.5	...
	High Asp.	1.3	0.9	...
7	Normal	1.5	0.7	...
	Double	2.7	0.7	...
	High Asp.	2.1	0.5	...

[a] High-aspiration.

TABLE IV
Deoxynivalenol Concentrations in Straight-Grade Flours Expressed as Percentages of the Concentrations in Uncleaned (UCW) or Cleaned (CW) Wheat

	Cleaning Method									
	Normal		Double		High Aspiration		Washer		Mean	
Lot	UCW	CW	UCW	CW	UCW	CW	UCW	CW	UCW	CW
1	55	70	52	59	58	66	53	60	54	64
2	24	39	23	53	35	104	28	54	28	62
3	41	88	31	76	35	58	44	98	38	80
4	50	76	67	97	55	77	62	85	59	84
5	33	61	15	41	45	81	67	92	40	69
Mean	41	67	38	65	46	77	51	78	44	72
SD	12	18	21	22	11	18	15	20	13	10

TABLE V
Loaf Volumes and Deoxynivalenol Concentrations of Breads Baked with Straight-Grade Flours from the Miag Multomat Mill—Means of Flour Cleaning Methods

Lot	Loaf Vol[a] (cm^3)	Deoxynivalenol Concentrations			
		Flour (ppm)	Bread[b] (ppm)	% Change From Flour	
				Mean[c]	Range
1	957	0.35	0.28	−19	−5 to −30
2	937	0.22	0.13	−42	−36 to −47
3	952	0.47	0.34	−27	−20 to −34
4	941	0.88	0.70	−21	−10 to −26
5	886	2.03	2.04	+0.6	+13 to −2
6	923	0	0		
7	910	0	0		

[a] Adjusted to 11% protein (Finney and Yamazaki 1967).
[b] Corrected for nonflour ingredients.
[c] Overall mean = −22 (SD 15).

TABLE VI
Deoxynivalenol Concentrations of Crumb and Crust of Breads Baked with Straight-Grade and Whole Wheat Flour

Lot	Flour Type	Deoxynivalenol Concentrations		
		Flour (ppm)	Bread[a] (% Change From Flour)	
			Crumb	Crust
4	Straight grade[b]	1.1	−4	0
4	Whole[c]	1.6	+12	+5
5	Straight grade	3.2	+25	+24
5	Whole	5.8	−7	+2

[a] Corrected for nonflour ingredients.
[b] Allis-Chalmers experimental mill.
[c] Weber hammer mill.

utilizes storage proteins, removes cell walls, and damages starch granules in heavily infected grain. Apparently such kernels tend to be fractured by the first and second break rolls to provide fine particles (flour) and particles of a size susceptible to reduction to flour by the first reduction rolls (Fig. 2). Although gross mill stream yields from infected grain did not appear to differ significantly from those milled from normal grain, it is possible that certain highly infected particles fractured abnormally, thus altering their particle distribution.

Baking Tests

Only flours from severely infected wheat (lot 5) produced loaves with slightly decreased volumes and less than satisfactory crumb color (Table V). Also, only lot 5 flour baking quality was affected by the method used to clean the wheat before milling. Loaves from lot 5 flours had crumb color that was questionable when wheat was cleaned by normal and high aspiration methods, questionable to satisfactory when cleaned by the double method, and unsatisfactory when cleaned by the washer method. The latter loaves had the lowest volume (841 cm^3, adjusted to 11% protein by the method of Finney and Yamazaki 1967) of all the loaves baked in this study.

Some baking tests indicated that breads had lower DON concentrations than flours, especially when DON concentration of flour was low (Table V), whereas other tests showed relatively little change in DON contents from flour to bread (Tables V and VI). However, none of our results indicated significant increases in DON concentrations during the breadmaking process. Young et al (1984) reported 118–189% increase in DON concentration of yeast doughnuts relative to flours, possibly due to enzymatic conversion of some precursor into DON.

A further baking test with straight-grade and whole wheat flours showed that DON concentrations of crumb and crust were not significantly different (Table VI). Apparently the higher temperature in crust compared to crumb has little or no effect on DON concentration.

Previously, Finney (1954) reported that flour from wheat containing 10% scab gave loaves with slightly reduced volumes and questionable crumb color and grain. Infected wheats with less than 10% scab were not included in that study. The present study suggests that flours from wheats containing up to about 3% scab will probably not noticeably decrease bread baking quality. The presence of DON in the bread is, of course, an undesirable quality because of its known toxicological effects in animals (Ueno 1983). The U.S. Food and Drug Administration has advised that wheats containing more than 2 ppm should not be used for milling and that flours containing more than 1 ppm should not be used for food products (Anonymous 1982).

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